The Cell - Transport Mechanisms and Cell Permeability

**Purpose:**

1. Define diffusion, osmosis, selective permeability, osmotic pressure, active and passive transport, concentration gradient, hypertonic solution, isotonic solution, hypotonic solution, crenation, and hemolysis.

2. Understand the relationship between rate of diffusion and molecular weight, and between selective permeability and molecule size.

3. State all reagents used and their purposes.

4. Explain all experimental results. For example, give reasons for weight gain in some sacs but not all.

**Material:**

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| 1. Four [dialysis bags](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/dbagstrings.jpg) |
| 2. Eight pieces of [twine/string](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/dbagstrings.jpg)    (two/bag) |
| 3. Four 250 ml beakers |
| 4. One funnel |
| 5. One test tube rack with six test     tubes |
| 6. Test tube forceps/clamp |
| 7. Wax pencils |
| 8. Flask containing 40%     dextrose/glucose |
| 9. Flask containing 10% NaCl     (sodium chloride) solution |
| 10. Flask containing 0.1% Starch |
| 11. Benedict's solution in dropper       bottle |
| 12. Silver nitrate in brown storage       bottle |
| 13. Iodine (also IKI) solution in       dropper bottle |
| 14. Flask containing distilled water |
| 15. 25-ml Graduated cylinder |
| 16. Top loading balance |
| 17. Boiling water baths (beaker        filled with water on a hot plate) |

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| **Procedure:** |
| 1. Locate all materials.  Some of them are available at your table while others are located at the instructor's desk or at the back of the laboratory (the hot plates, for instance).  Know where **everything** is located as this will make the lab run smoothly. |
| 1. Obtain the four dialysis bags (each member of the group should get one).  Wet the bag and [tie off one end](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/TIEDDBAG.JPG).  Be careful not to pull too tight as this might tear the bag, but be sure it is very secure.  If the bag dries, wet it again and rub (gently) it between your index and thumb to open the other end.  (This can be tricky - the key is PATIENCE.). |
| 1. (dialysis bag #1) - Once the bag is opened [place the tip of the funnel into the bag](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/AddSolntoBag.jpg) and **add 8 milliliters (ml) of 40% dextrose** using the graduated cylinder.  Be sure to do this over the laboratory sink.  [Close the opened end by tying it](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/preparedbag.jpg) with string (again, be sure it is secure and there are no leaks).  Rinse off any excess dextrose on the outside of the bag and pat dry with paper towels. |
| 1. Repeat step 3 for each of the solutions:   (2) 10% NaCl (3) 0.1% Starch (4) Prepare another bag with 40% dextrose. |
| 1. Once all bags have been patted dry, [**weigh each one**](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/weighbag.jpg) using the top loading balance. Use the **TARE** button to reset the balance.  Each of the front four laboratory tables have a balance. If you do not have one at your table, simply walk over to a table that does and weigh all your bags.  **Be sure to record the weight of each bag in your chart (see below).** |
| 1. Obtain and label four beakers (1-4).  Place one prepared dialysis bag into each beaker.[**Be sure to note**](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/DBagAnalysis.htm) **which dialysis bag was placed in which labeled beaker**. |
| 1. Add distilled water to all the beakers **EXCEPT** the one containing the second bag of 40% dextrose.  [**Add enough water until the bag is completely submerged**](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/baginbeaker.jpg). To the beaker with the second bag of 40% dextrose add 40% dextrose.  Again, add enough dextrose so the bag is completely submerged. **Be sure to record the time** at which the prepared bags were placed into the beakers of water (and dextrose). Your set up should like the [illustration provided](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/SetupDialysisIllustration.htm). |
| 1. Leave bags in beakers for 45 minutes.  You may continue with the other experiments during this time. |
| 1. At the end of the 45 minute period, remove bags one at a time and pat dry to remove excess water.  Weigh each bag and record the weight. **Did all the bags maintain their weight?  Why?  or Why not?**   **DO NOT discard the water in the beakers.**  They will be tested using the procedure below. |
| 1. Obtain the test tubes, test tube rack, marker, tape (found at instructor's desk), test tube clamp, and [testing reagents](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/dialysisreagents.jpg). |
| 1. Separate the test tubes into pairs.  Label the test tubes as [indicated](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/DBagAnalysis.htm). Be sure to keep all respective beakers and tubes together to avoid any "mix ups". |
| 1. Take the dialysis bag containing 40% dextrose and carefully remove the string from one end.  Using a pipette, [remove about 5 ml of the solution](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/dbagsample.jpg).  Add this [sample to the test tube](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/dbagtesttube.jpg) marked "Control D" and place in the test tube rack.  Add 5 ml of Benedict's solution to the test tube. |
| 1. Take 10 ml of water from the beaker (that had the dialysis bag containing 40% dextrose) and add it to the test tube labeled "Test D" and place in the test tube rack.  Add 5 ml of Benedict's solution to the test tube. |
| 1. Take both tubes and the test tube forceps to the boiling water baths located in the back of the laboratory class.  [Attach the forceps to one of the test tube](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/forceps.jpg) and [place carefully into](http://www.usi.edu/science/biology/mkhopper/hopper/BIOL2401/LABUNIT1/01Ex5CellTransp/CellTransport%20Images/boilingbath.jpg) the boiling water bath.  Repeat this step for the other tube.  Boil both tubes for about three minutes. |
| 1. After the boiling period carefully remove tubes with the forceps and place into test tube rack.  Return to your table.  **The solution in the tube labeled "Control D" will undergo a color change.  The change from blue to orange or red orange indicates the presence of glucose/dextrose.  Did the tube labeled "Test D" change colors?  If so, what does this mean?** |
| 1. Obtain the dialysis bag containing 10% Na Cl and the respective beaker.  Remove 5 ml of the solution from the bag (as you did in step 12) and add it to the test tube labeled "Control N" and place in the test tube rack.  Take 10 ml of water from the beaker and add it to the test tube labeled "Test N" and place it in the test tube rack.  Add, carefully, three drops of silver nitrate (It is located at the instructor's desk.) into each tube.  **The solution in the tube labeled "Control N" will form a white precipitate upon the addition of the silver nitrate.**  **Did the tube labeled "Test N" form a white precipitate?  If so, what does this mean?** |
| 1. Obtain the dialysis bag containing 0.1% starch and the respective beaker.  Remove 5 ml of the solution from the bag (as you did in step 12) and add it to the test tube labeled "Control S" and place in the test tube rack.  Take 10 ml of water from the beaker and add it to the test tube labeled "Test S" and place it in the test tube rack.  Add, carefully, five drops of IKI (iodine) into each tube.  **The solution in the tube labeled "Control S" will change into a navy blue (or even black) color upon the addition of the IKI.**  **Did the tube labeled "Test N" form a white precipitate?  If so, what does this mean?** |
| 1. **What was the purpose of placing a dialysis bag containing 40% dextrose in a beaker of 40% dextrose?  Do you need to test any of the contents (in the beaker and/or bag)?  Why or Why not?  Was there a significant change in the weight of the bag after the 45 minute period?  Can you explain?** |
| 1. After you have completed this experiment, drain the dialysis bags into the laboratory sink and discard them (and the string) in the trash can.  Wash (soap and test tube brush are available at the faucet) all test tubes and clean the table. |

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| **Data Table** | |  | |
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| **Beaker** | | **Contents of sac**  **(5-8 ml)** | | **Initial weight**  **(g)** | | **Final weight**  **(g)** | **Weight Change:**  **+ or – (g)** | **Tests on Beaker fluid** | **Tests on Sac Fluid** | **Observations** |
| Beaker 1  (distilled water) | | 40% glucose solution | |  | |  |  | Benedict’s test: **(test D)** | X |  |
| Beaker 2  (distilled water) | | 10% NaCl solution | |  | |  |  | AgNO3  Test N | AgNO3  **Test N (control)** |  |
| Beaker 3  (distilled water) | | .1% Starch  Solution | |  | |  |  | KI solution:  **Test S** | KI solution  **Test S**  **(Control)** |  |
| Beaker 4  (40% glucose solution) | | 40% glucose solution | |  | | sucrose solution containing dye |  | X | **(test D Control)** |  |

**Conclusion:**

* 1. **Did all the bags maintain their weight?  Why?  or Why not?**
  2. **The solution in the tube labeled "Control D" will undergo a color change.  The change from blue to orange or red orange indicates the presence of glucose/dextrose.  Did the tube labeled "Test D" change colors?  If so, what does this mean?**
  3. **The solution in the tube labeled "Control N" will form a white precipitate upon the addition of the silver nitrate.**  **Did the tube labeled "Test N" form a white precipitate?  If so, what does this mean?**
  4. **The solution in the tube labeled "Control S" will change into a navy blue (or even black) color upon the addition of the IKI.**  **Did the tube labeled "Test N" form a white precipitate?  If so, what does this mean?**
  5. **What was the purpose of placing a dialysis bag containing 40% dextrose in a beaker of 40% dextrose?  Do you need to test any of the contents (in the beaker and/or bag)?  Why or Why not?  Was there a significant change in the weight of the bag after the 45 minute period?  Can you explain?**